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First records of *Inocybe melleiconica* and *I. pararubens* for Northern Europe with a new variety from the alpine zone of the Scandinavian mountains

Jukka Vauras^{1,*} and Ellen Larsson²

- ¹ Biological Collections of Åbo Akademi University, Herbarium, FI-20014 University of Turku, Finland
- ² University of Gothenburg, Department of Biological and Environmental Sciences, Gothenburg Global Biodiversity Centre, Box 461, SE-40530 Göteborg, Sweden

*Corresponding author:

jukvau@utu.fi

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Abstract

Inocybe melleiconica and I. pararubens are reported as new for Northern Europe, from the alpine zone of Scandinavia. I. melleiconica is rather common on rich and more calcareous soils and usually associated with Salix herbacea. I. pararubens seems to be a rather rare species in the alpine zone and only encountered on calcareous soils on wet ground, associated with Salix reticulata and S. herbacea. We describe I. pararubens var. padjelantae to accommodate the alpine collections of I. pararubens. This taxon is also identified from Canada, arctic tundra of Keewatin. I. pararubens var. pararubens is recorded as new for Sweden, where it was found associated with Tilia cordata, on rich calcareous soil on the west coast of the country. I. castaneicolor is identified as a later synonym of I. pararubens. The spores of the alpine collections of both species treated here were found to be clearly broader than the ones given in the original descriptions.

Introduction

During the last ten years several excursions in different regions of the alpine zone of Fennoscandia have been made to explore the fungal diversity. Among the visited sites are e.g. the Abisko Mountains, Padjelanta National Park and Vuoggatjålme in Sweden, and Kilpisjärvi and Utsjoki areas in Finland. It is known that the highest plant diversity in the arctic-alpine zones is connected to areas with calcareous soils. This is also the case for the fungi, so localities known to host a high vascular plant diversity indicating calcareous soils were targeted.

One of the major groups of Agaricales associated in the alpine zone with dwarf *Salix*, *Dryas octopetala*, *Bistorta vivipara* and other ectomycorrhizal forming plants is *Inocybaceae*. This family has been in focus of our collecting and we have been collected hundreds of specimens. The morphological and molecular studies have revealed a large and previously partly unknown hidden species diversity from these regions. Many species new to science have been described and several ambiguous names have been sorted out. Also, species described from the alpine zone in Europe by e.g. Favre (1955), Kühner (1988) and Bon (1997) have been identified to occur in Fennoscandia (Larsson et al. 2014, 2017, 2018, Cripps et al. 2019).

Some species of Inocybaceae growing in deciduous forests on calcareous soils further south in Fennoscandia and Europe, such as I. grammata Quél. and Inosperma erubescens (A. Blytt) Matheny & Esteve-Rav., are also encountered in the arctic-alpine zones on calcareous to ultra basic soils, usually associated with Dryas octopetala and Salix reticulata. Here we report findings of I. pararubens Carteret & Reumaux originally described from Fagus forest in France, and the here identified synonym I. castaneicolor La Rosa, Bizio, Saitta & Tedersoo decribed from South Italy associated with Quercus suber from the alpine zone in Scandinavia. We also report I. melleiconica, originally described from Canada and associated with *Tsuga canadensis*, from the alpine zone of Scandinavia. Emended macro- and micromorphological descriptions and ecology for the two species are provided based on our specimens.

Material and Methods

Sequences from the complete internal transcribed spacer (ITS barcode) region of the nuclear ribosomal DNA were generated for most specimens. DNA extractions, PCR reactions, primers used, and sequencing were performed as described in Larsson et al. (2018). The ITS of the target species were then compared to those in GenBank (Clark et al. 2016) and the UNITE database (Kõljalg et al. 2013) to see for the best match. Results from the blast searches show that one of our target species were 100% identical with the ITS of the isotype of I. melleiconica Grund & Stuntz (NG057290). The other target species were 100% identical with the holotype sequence of I. castaneicolor (KY213954) and with the holotype sequence of I. pararubens Carteret & Reumaux (MN954310) newly generated in this study.

Morphological comparison with the species descriptions confirm the identity and also that *I. castaneicolor* is a later synonym of *I. pararubens*.

Macroscopic characters were noted, and cross-sections were drawn or photographed. The colour codes refer to Cailleux (1981) or Küppers (1981). Microscopic examination was completed on dried specimens in 10% NH4OH, typically at ×1250 magnification. The size of basidiospores is given as length \times width. The number of spores measured is given for both taxa. Means are placed between range limits. Q denotes the quotient of length divided by width for each spore, which are averaged for Qm. Measurements of basidia are for length \times width without sterigma included. Measurements of cystidia are given as length \times width, without inclusion of crystals at the apex.

Taxonomy

Inocybe melleiconica Grund & D.E. Stuntz - Figs. 1-5

Pileus 1.6–4.0 cm in diam, when young hemisphaerical to conico-convex, later conico-convex to



Fig. 1. *Inocybe melleiconica, Vauras 32532F.* Fruiting bodies in alpine zone in Sweden, Arjeplog. Photo: J. Vauras.



Fig. 2. Inocybe melleiconica, Larsson & Vauras 32894F. Fruiting bodies in alpine zone in Sweden, Strömsund. Photo: J. Vauras.







Fig. 4. Inocybe melleiconica. Microscopical characters (isotype). For symbols, see Fig. 3.

plano-convex, without or with broad, low umbo, but sometimes with prominent umbo, margin deflexed, straight to reflexed, pale brown (Cailleux 47P, 20M), greyish yellow-brown (69M, 70M), to pale brownish grey (Küppers S20Y30M10), centre cream, watery yellow (S20Y99M20), pale brown to brown, smooth around centre, towards margin fibrillose, fibrils at margin sometimes darker, partly subsquamulose, silky shiny, margin often breaking up and sometimes slightly appendiculate; when young sometimes with silvery-white velipellis. *Lamellae* moderately crowded, up to 6 mm wide, subventricose, narrowly adnate, first pale grey, beige-grey (91K), then brownish grey, grey-brown (70M-N), pale brown to brown, edge concolorous, fimbriate. *Stipe* $2.2-5.0 \times 0.2-0.65$ cm, equal to slightly clavate, base often bulbous, but rarely marginately bulbous, up to 0.7 cm wide; whitish, slightly yellowish to reddish,



Fig. 5. Locality of *Inocybe melleiconica* in Sweden, Jukkasjärvi, Abisko mountains, near Latnjajaure 19.VIII.2013. Photo: J. Vauras.

base white, white-pruinose to the base, longitudinally striate, lower part often more hairy-pruinose. *Cortina* not observed. *Context* in pileus whitish, sometimes partly brownish, in stipe whitish, slightly reddish, brownish to orange, whitish at base, shiny. *Smell* spermatic.

Spores $(7.7-)8.2-9.4-10.5(-11.2) \times (5.0-)5.2-$ 5.7–6.3(–6.5) μ m, range of mean values 9.0–10.0 × 5.4–6.0 μ m, Q = (1.4–)1.45–1.66–1.8(–1.9), range of mean Q -values 1.56-1.77 (140 spores from 7 collections); smooth, subamygdaliform, mostly with subconical apex, yellow brown. Basidia (22-)24- $29-34(-35) \times 8-10-11 \mu m$, clavate, mainly 4-spored (60 basidia from 3 collections). Pleurocystidia $(46-)50-62-77(-86) \times (12-)14-17-21(-23) \ \mu m \ (120)$ pleurocystidia from 7 collections), subfusiform to subutriform, often with a short to long pedicel, thickwalled, with up to 4 µm thick, colourless to pale yellow wall, mostly with scarce to abundant crystals; frequent. Cheilocystidia somewhat similar to pleurocystidia but more variable and partly with rounded base, several with yellow-brown contents, frequent, $(33-)38-55-70(-83) \times (11-)12-16-20(-24) \ \mu m \ (n$ = 65); paracystidia moderately scarce, pyriform to clavate, $(13-)15-20-26(-29) \times 8-11-15(-16) \mu m$ (n = 52). *Caulocystidia* descending to base of stipe, (26-)36-54-74(-85) × (10-)12-16-20(-24) µm (n = 108), more variable than pleurocystidia, at base often cylindrical; cauloparacystidia mostly clavate, pyriform or oval, $(11-)15-21-28(-32) \times 7-11-14(-16) \mu m$ (n = 30). *Clamp connections* present in all tissues.

Isotype of *Inocybe melleiconica* Grund & D.E. Stuntz. CANADA, Nova Scotia, Kentville, Kings Co., 4.X.1966, *H. Stewart, DG 1906* (WTU). 2 halves of fruit bodies. *Spores* (8.2–)8.3–9.6–10.5(–11.5) × (4.4–)4.5–4.9–5.1(–5.5) µm, Q = (1.6–)1.65–1.97–2.25(–2.3) (n = 20), smooth, subamygdaliform. *Basidia* 4-spored, e.g. 33×9 µm. *Pleurocystidia* 57–63–68 × 17–20–25 µm, subfusiform to subutriform, mostly with a pedicel, thick-walled, with up to 4 µm thick, almost colourless wall, mostly with scarce to abundant crystals; frequent. *Cheilocystidia* 39–55–61 × 10–18–22 µm (n = 5), more variable than pleurocystidia. *Caulocystidia ia* descending to base of stipe, 29–45–59 × 11–15–30 µm (n = 9), more variable than pleurocystidia, rather cylindrical at base.

Discussion Troms. Storfjord, Skibottsdalen, N of Rieppejavri lake, 24.VIII.1995 J. Vauras 10640F (TUR-A). SWEDEN. Inocybe melleiconica described in this study has Härjedalen, Tännäs, Svansjökläppen, 16.VIII.2006, mostly medium-sized fruiting bodies, pale brownish grey to brown pilei, totally pruinose pale stipe, smooth, subamygdaliform spores which are on average 9-10 µm long and 5.4-6 µm broad, and growing in alpine zone with dwarf willows. The species was described first from Canada, Nova Scotia, growing under hemlock (Tsuga canadensis). Our material fits

E. Larsson 95-06 (GB). Jämtland, Frostviken, Domprosten, 24.VIII.2019 J. Olsson & J. Vauras 32913 (TUR-A), Raavre, 23.VIII.2019 E. Larsson & J. Vauras 32893F, 32894F (TUR-A, GB). Åsele lappmark, Vilhelmina, Klimpfjäll area, Frimtstjakke, 22.VIII.2019 J. Vauras 32885 (TUR-A), Lasterfjället, Tjårronjunjes, 19.VIII.2019 J. Vauras 32833 (TUR-A), Marsfjällen, Fiehteres, 21.VIII.2019 J. Vauras 32866F (TUR-A, GB). Lycksele lappmark, Arjeplog, Àhkáris, 14.VIII.2108 J. Vauras 32532F (TUR-A, GB), Nuortta Kráhpasvarre, 12.VIII.2108 J. Vauras 32500 (TUR-A, GB), J. Vauras, J.B. Jordal & E. Larsson 96-18 (GB). Lule lappmark, Jokkmokk, Padjelanta National Park, Arralåbbdå 11.VIII.2016 E. Larsson 40-16 (GB), Vielggisbákte, 12.VIII.2016 J. Vauras 31482F (TUR-A, GB), Tuottar, 13.VIII.2016 J. Vauras 31500F (TUR-A, GB), N side of Slahpejávrre, 14.VIII.2016 J. Vauras 31512 (TUR-A, GB), J. Vauras 31523F (TUR-A, GB), near Njoammeljávrre, 18.VIII.2016 J. Vauras 31580 (TUR-A, GB), E. Larsson 228-16 (GB), Svártinjunjes 19.VIII.2016 E. Larsson 259-16 (GB). Torne lappmark, Jukkasjärvi, Abisko, near Latnjajaure, 16.VIII.2013 E. Larsson & J. Vauras 29910F (TUR-A), P.-A. Moreau, J. Vauras & E. Larsson 35-16 (GB). 19.VIII.2013 P.-A. Moreau (J. Vauras 29950) (TUR-A), P.-A. Moreau, J. Vauras 29946F & E. Larsson 103-13 (GB, TUR-A). 20.VIII. 2013 P.-A. Moreau (J. Vauras 29955) (TUR-A), 21.VIII.2013 E. Larsson 159-13, P.-A. Moreau & J. Vauras (GB, TUR-A), E. Larsson & J. Vauras 29972 (TUR-A), 22.VIII.2013 E.

NORWAY.

SPECIMENS

EXAMINED:

Larsson 183-13, P.-A. Moreau & J. Vauras (GB, TUR-A). Inocybe melleiconica is known from several localities of the Scandinavian Mountains in Sweden and Norway. All collections were found in alpine belt, on rich, ± calcareous, not too wet ground, mainly amongst Salix herbacea, at elevations of 700-1100 m a.s.l. Sometimes also Salix glauca, S. reticulata or Dryas octopetala were observed near I. melleiconica. The herbs Bistorta vivipara and Sibbaldia procumbens are often present at the growth sites, as well as Equisetum variegatum. All our Nordic specimens were collected in mid- to late-August.

well to the original description by Grund & Stuntz (1968), but in our collections the spores are broader (4.5-5.5 µm in the original description), and pleurocystidia are not so thick-walled (3.5-6 µm in the original description). The same situation about the spores was observed when comparing spore sizes of I. semifulva Grund & D.E. Stuntz. They were broader (5.5-6.3 µm) in Finnish sequenced specimens (Vauras 2018) than in original material from Nova Scotia, Canada, 4.5-5(-5.5) µm (Grund & Stuntz 1981). Our drawing of the microscopical characters of the isotype is given in Fig. 4. ITS sequence data of our alpine collections and the isotype of I. melleiconica (NG057290) are identical. According to Matheny (2017) I. melleiconica is phylogenetically a part of I. hirtella group.

Pale fruitbodies of I. melleiconica could be misidentified as I. ochroalba Bruylants. This species has a wide distribution in the Nordic countries but seems to grow only rarely in the alpine zone. It is paler, has somewhat smaller spores and shorter, clavate to almost subglobose cystidia (Jacobsson & Larsson 2012).

Inocybe pararubens Carteret & Reumaux var. padjelantae E. Larss. & Vauras var. nova

- Figs. 6-9

MycoBank no.: MB 834179

DIAGNOSIS: Fruiting bodies small to medium-sized, with dark brown, brown to orange-brown pilei 1-3 cm in diam, totally pruinose pale stipes measuring $1.0-2.7 \times 0.2-0.6$ cm, with base often bulbous but not marginately bulbous, spores smooth, rather large, on average 10.5-11.3 µm long and 6.8-7.3 µm broad,



Fig. 6. Inocybe pararubens var. padjelantae, Vauras 31591F. Fruiting bodies in alpine zone in Sweden, Jokkmokk. Photo: J. Vauras.

some of them often with apical papilla. Differs from the specimens collected in deciduous forests (*I. pararubens* var. *pararubens*) by broader spores and herewith smaller Q-values of spores (range of mean values 1.44–1.59), and in the comparison of ITS sequence data by one substitution. In alpine zone on calcareous, wet ground with dwarf willows.

TYPUS: SWEDEN. Lule lappmark, Jokkmokk, Padjelanta National Park, NE side of Sårjåsjávrre, SW slope of Sårjåsthjåhkkå, gently sloping alpine heath, at small brook with *Salix reticulata* and *S. herbacea*, on mosses, on calcareous soil, alt. ca 850 m, 17.VIII.2016 *J. Vauras 31552F* (TUR-A 204347 – holotypus, GenBank Acc. No. MN994329; GB, Herb. D. Bandini – isotypi).

ETYMOLOGY: *padjelantae* (gen.) refers to Padjelanta National Park in Jokkmokk commune in Sweden, where nearly all our collections were found.

PILEUS 1.0–3.0 cm in diam, when young hemisphaerical to conico-convex, later plano-convex, often with broad, low umbo, margin somewhat inflexed when young, then straight to deflexed; centre dark brown, blackish, blackish-brown, reddish-brown, pale brown, outwards dark brown, brown, greyish-brown to orange-brown, at margin sometimes pale greyish, smooth around centre, silky shiny, towards margin slighty fibrillose, fibrils sometimes darker, margin slightly breaking up, no velipellis seen. Lamellae subdistant, up to 4 mm wide, ventricose, narrowly adnate, first pale grey, then yellowish grey, greyish-brown, later yellow-brown to brown; edge pale to concolorous, fimbriate. Stipe $1.0-2.7 \times 0.2-0.6$ cm, cylindrical to clavate, base often bulbous but not marginately bulbous, up to 0.7 cm wide; first whitish, then slightly yellowish to brownish, sometimes partly brown to blackish-brown, white-pruinose to the base, longitudinally striate, lower part somewhat white-felted. Cortina not observed. Context watery whitish to yellowish, shiny. *Smell* indistinct to slightly spermatic. Spores $(9.4-)10.0-10.9-12.0(-13.1) \times (6.1-)6.4-$ 7.2-7.9(-9.4) µm, range of mean values $10.5-11.3 \times$ 6.8-7.3 μm, Q = (1.35-)1.4-1.53-1.7(-1.75), range of mean Q-values 1.44-1.59 (100 spores from 5 collections); smooth, ± subamygdaliform, mostly with al-



Fig. 7. *Inocybe pararubens var. padjelantae, holotype.* Fruiting bodies in alpine zone in Sweden, Jokkmokk. Photo: J. Vauras.

most rounded to subconical apex, some with apical papilla, yellow brown. Basidia $24-29-38 \times (9-)10-$ 11-13(-14) µm, clavate, mainly 4-spored (39 basidia from 5 collections). Pleurocystidia (55-)61-74-90(-96) × (12-)14-18-23(-31) μm (98 pleurocystidia from 5 collections), subfusiform to subutriform, often with a pedicel, thick-walled, with up to 4 μ m thick, pale yellowish wall, with scarce to abundant crystals; frequent. Cheilocystidia somewhat similar to pleurocystidia but more variable and partly with rounded base, several with yellow-brown contents, frequent, $(40-)52-64-77(-86) \times (13-)14-17-22 \ \mu m$ (n = 40); paracystidia fairly abundant, oval, pyriform to clavate, $16-21-26 \times 8-11-14(-15) \mu m$ (n = 24). Caulocystidia descending to base of stipe, (35- $)46-63-80(-92) \times (14-)15-17-20 \ \mu m \ (n = 20), more$ variable than pleurocystidia, at base often rounded; cauloparacystidia mostly clavate, pyriform or oval, $12-16-24 \times 10-13-15 \ \mu m$ (n = 14). Clamp connections present in all tissues.

Collection of *I. pararubens* var. *pararubens* from southern coast of Sweden, Bohuslän, Tanum, Lindö, 7.IX.2017 *E. Larsson 260-17* (GB, TUR-A): *Spores* $(10.0-)10.3-11.4-12.0(-12.8) \times (5.8-)5.9-6.4-7.0(-$

7.7) μ m, Q = 1.6–1.76–1.95(–2.0) (n = 20). *Pleurocys-tidia* (54–)55–62–66(–68) × 14–16–17 μ m (n = 20).

SPECIMENS EXAMINED: CANADA. North West Territories, Keewatin, 16.VIII.1971 *E. & M. Ohenoja* 1971-36 (OULUF027325); SWEDEN. Lule lappmark, Jokkmokk, Padjelanta National Park, Svártinjunjes, 19.VIII.2016 *E. Larsson 251-16* (GB, TUR-A), *E. Larsson 252-16* (GB), *J. Vauras 31591F* (TUR-A, GB); Sårjåstjåhkkå, 17.VIII.2016 *E. Larsson 204-16, 209-16* (GB, TUR-A), *J. Vauras 31552F, 31553F, 31557F* (TUR-A, GB); Unna Duvgge, 15.VIII.2016 *E. Larsson 160-16* (GB, TUR-A). Pite lappmark, Arjeplog, Ikesvagge, SW side of Gabsjetjåhkkå, 13.VIII.2018 *J. Vauras, J.B. Jordal & E. Larsson 112-18* (GB).

Inocybe pararubens var. *padjelantae* is known from Sweden from a few localities in Padjelanta National Park and the Vuoggatjålme area on the Scandinavian Mountain Range, and from one locality at the coast of the North Sea. Here we also confirm the species from Canada, collected in low arctic tundra in the District of Keewatin. All alpine collections from Sweden were found on rich, calcareous and rather wet ground, amongst *Salix reticulata* and/or



Fig. 8. *Inocybe pararubens var. padjelantae, holotype.* Microscopical characters and cross sections of fruiting bodies. For symbols, see Fig. 3.

S. herbacea, at elevations of about 800–850 m a.s.l. The herb *Bistorta vivipara* is often present at the growth sites, as well as *Equisetum variegatum*, and sometimes *Thalictrum alpinum* and *Saxifraga oppositifolia*. The species is clearly demanding and rare. All alpine specimens were collected in mid-August. In the comparison of ITS sequence data, the alpine collections differ in only one substitution from the type and specimens collected in deciduous forest.

Discussion

Inocybe pararubens var. padjelantae is characterized in our alpine material by small to medium-sized fruiting bodies, with dark brown, brown to orange-brown pilei, totally pruinose pale stipes, smooth, rather large, on average $10.5-11.3 \mu m \log p$ and $6.8-7.3 \mu m$ broad spores, some of them often with apical papilla, and growing in alpine zone with

dwarf willows. The species was first described from Northern France, growing under Fagus sylvatica (Carteret & Reumaux 2012). In the paper I. pararubens was included in the section Lilacinae. Later the species was published from Southern Italy, Sicilia, as I. castaneicolor, growing under Quercus suber (La Rosa et al. 2017). The species was then placed in section Splendentes based on the presented phylogeny where the species got a place in between I. phaeoleuca Kühner and I. tjallingiorum Kuyper. Our material fits rather well the original descriptions, but the spores are clearly broader than in the original descriptions, 5-6(6.5) µm in Carteret & Reumaux (2011) and (5)5.3-5.8(6.2) µm in La Rosa et al. (2017). Therefore the Q-values of the spores reported here are much smaller. Also the pleurocystidia are on average larger in our alpine material. Further, in our material the stipes of fruit bodies were not marginately bulbous, as reported from Italy by La Rosa et al. (2017).

In alpine areas *Inocybe pararubens* var. *pad-jelantae* could be mixed macroscopically with *I. lei-*



Fig. 9. Type locality of *Inocybe pararubens* var. *padjelantae* in Sweden, Jokkmokk, Padjelanta National Park 17.VIII.2016. Photo: J. Vauras.

ocephala. However, this species has smaller spores $(8.8-10.0-11.4 \times 5.6-6.2-7.0 \ \mu\text{m})$ and pleurocystidia $(53-64-81 \times 12-16-21 \ \mu\text{m})$, which are mainly with many small crystals at apex (Larsson et al. 2014).

Based on morphology alone, because of the different values of the spores, particularly the width and the Q-values, compared with the types of both treated species in this article, it would not have been possible to identify our collections. Thanks to the molecular methods, comparison of ITS sequence data, we have succeeded to identify them and have not described two new species in vain.

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